3 QUALITY CONTROL OF CRITICAL REAGENTS AND SUPPLIES

The following reagents/kits/supplies will be quality control tested as indicated below when a new lot is received into the laboratory.

3.1 PCR- Based System Kits

3.1.1 Upon receipt and before use, each new lot of PCR-based kits will be quality control tested to ensure the integrity of the PCR reagents that are provided with the kit. **If the same lot of the PCR-based system kits is received for several shipments, it is not necessary to quality control test the kits again until a new lot is received into the laboratory**.

3.1.2 Amplify and type two samples that have been extracted of known DNA type, one negative amplification blank, and one positive amplification control (K562 or GM9947A Cell Line) per lot of new kits. Follow the amplification, product gel, and typing gel procedures outlined in the Commonwealth of Virginia Division of Forensic Science Forensic Biology Section Procedure Manual, Section III, Fluorescent Detection PCR-Based STR DNA Protocol PowerPlex® 16 BIO System. **Record the loading position of the samples on the worksheets used for casework analysis.**

3.1.3 Once the gel images have been sized, provided the expected types and intensities are obtained (i.e., known samples and positive amplification control) and the negative amplification blank does not produce results, the new lot of PCR- based kits will be considered to have passed the quality control verification and may be put on-line for routine use. **The quality control verification of the PCR-based kits does not need to be sized by an independent sizer**.

3.1.5 Once the new lot of PCR-based kits has passed the quality control verification, the kits will be labeled with the quality control date and the expiration date. Refer to the Commonwealth of Virginia Division of Forensic Science Forensic Biology Section Procedure Manual, Section III, Fluorescent Detection PCR-Based STR DNA Protocol PowerPlex® 16 BIO System for the PCR- based system kit expiration date. All worksheets and paperwork associated with the quality control test will be maintained in a ring binder or properly labeled file.

NOTE: This quality control test can be done in conjunction with the quality control testing of the PAGE Plus™ gel Solutions, or AmpliTaq™ Gold DNA polymerase.

3.2 PAGE Plus™ Gel Solutions

The PAGE Plus™ Gel Solution (acrylamide gel) is used to separate amplified STR alleles. This procedure is designed to verify the integrity of each lot of PAGE Plus™ Gel Solution before it is used to separate the amplified STR alleles.

3.2.1 Prepare a 6.0% PAGE Plus™, 0.4 mm acrylamide gel in 1X TBE buffer. Refer to the Commonwealth of Virginia Division of Forensic Science Forensic Biology Section Procedure Manual, Section III, Fluorescent Detection PCR-Based STR DNA Protocol PowerPlex® 16 BIO System, for instruction on how to prepare the typing gel.

3.2.2 Using one negative amplification blank, and one positive amplification control (K562 or GM9947A Cell Line) per lot gel solution, follow the typing gel procedures outlined in the Commonwealth of Virginia Division of Forensic Science Forensic Biology Section Procedure Manual, Section III, Fluorescent Detection PCR-Based STR DNA Protocol PowerPlex® 16 BIO System. Prepare a sufficient volume of the allelic ladder so that two allelic ladders will be loaded into the acrylamide gel flanking the positive (K562 or GM9947A Cell Line) and negative amplification controls. **Record the loading position of the samples and requisite ladders on the worksheets used for casework analysis.**

3.2.3 Once the gel image has been sized, providing the correct result is obtained for the positive amplification control and all sample and allelic ladder bands are sharp and the amplification blank does not produce results, the new lot of PAGE Plus™ gel solution will be considered to have passed the quality control verification and may be put on-line for routine use. **The quality control verification of the PAGE Plus™ gel solution does not need to be sized by an independent sizer.**

NOTE: If a particular lot of gel solution is found to run faster or slower than expected and the gel solution provides sharp and clear results, it is not necessary to reject the quality control results. However, this information will be provided to the casework and Data Bank staff in order to adjust the running time for the particular lot of gel solution.

3.2.4 Once the new lot of PAGE Plus™ gel solution has passed the quality control verification, the stock solution (i.e., box or bottle) will be labeled with the quality control date and the expiration date. Refer to the Commonwealth of Virginia Division of Forensic Science Forensic Biology Section Procedure Manual, Section III, Fluorescent Detection PCR-Based STR DNA Protocol PowerPlex® 16 BIO System for the PAGE Plus™ gel solution expiration date. All worksheets and paperwork associated with the quality control test will be maintained in a ring binder or properly labeled file.

NOTE: This quality control test can be done in conjunction with the quality control testing of the PCR-based kits, AmpliTaq™ Gold DNA polymerase or PowerPlex®16 BIO Allelic Ladder or Internal Lane Standard 600 BIO.

3.3 AmpliTaq™ Gold DNA Polymerase

3.3.1 Each new lot of AmpliTaq™ Gold DNA polymerase will be quality control tested before used on casework or data bank samples to verify the activity of the enzyme. **If the same lot of AmpliTaq™ Gold DNA polymerase is received for several shipments, it is not necessary to quality control test the AmpliTaq™ Gold DNA polymerase again until a new lot is received into the laboratory.**

3.3.2 Amplify and type two samples that have been extracted of known DNA type, one negative amplification blank, and one positive amplification control (K562 or GM9947A Cell Line) per lot of new AmpliTaq™ Gold DNA polymerase. Follow the amplification, product gel, and typing gel procedures outlined in the Commonwealth of Virginia Division of Forensic Science Forensic Biology Section Procedure Manual, Section III, Fluorescent Detection PCR-Based

STR DNA Protocol PowerPlex® 16 BIO System. **Record the loading position of the samples on the worksheets used for casework analysis.**

3.3.3 Provided the expected types and intensities are obtained and the amplification blank does not produce results, the new lot of AmpliTaq™ Gold DNA polymerase will be considered to have passed the quality control verification and may be put on-line for routine use.

3.3.4 Once the new lot of AmpliTaq™ Gold DNA polymerase has passed the quality control verification, the AmpliTaq™ Gold DNA polymerase stock box will be labeled with the quality control date. Refer to the Commonwealth of Virginia Division of Forensic Science Forensic Biology Section Procedure Manual, Section III, Fluorescent Detection PCR-Based STR DNA Protocol PowerPlex® 16 BIO System for the AmpliTaq™ Gold DNA polymerase expiration date. All worksheets and paperwork associated with the quality control test will be maintained in a ring binder or properly labeled file.

NOTE: This quality control test can be done in conjunction with the quality control testing of the PCR-based kits, PAGE Plus™ gel solution, PowerPlex® 16 BIO Allelic Ladder or Internal Lane Standard 600 BIO.

3.4 PowerPlex® 16 BIO Allelic Ladder or Internal Lane Standard 600 BIO

3.4.1 Type one negative amplification blank and one positive amplification control (K562 or GM9947A Cell Line). Prepare a sufficient volume of the allelic ladder so that two allelic ladders will be loaded into the acrylamide gel flanking the positive (K562 or GM9947A Cell Line) and negative amplification controls. Follow the typing gel procedure outlined in the Commonwealth of Virginia Division of Forensic Science Forensic Biology Section Procedure Manual, Section III, Fluorescent Detection PCR-Based STR DNA Protocol PowerPlex® 16 BIO System. **Record the loading position of the samples on the worksheets used for casework analysis**.

3.4.2 Once the gel images have been sized, providing the expected types and intensities are obtained (i.e., positive amplification control) and the negative amplification blank does not produce results, the new lot of PowerPlex® 16 BIO Allelic Ladder or Internal Lane Standard 600 BIO will be considered to have passed the quality control verification and may be put on-line for routine use. **The quality control verification of the PowerPlex® 16 BIO Allelic Ladder or Internal Lane Standard 600 BIO does not need to be sized by an independent sizer**.

3.4.3 Once the new lot of PowerPlex® 16 BIO Allelic Ladder or Internal Lane Standard 600 BIO has passed the quality control verification, the stock box or bag will be labeled with the quality control date. Refer to the Commonwealth of Virginia Division of Forensic Science Forensic Biology Section Procedure Manual, Section III, Fluorescent Detection PCR-Based STR DNA Protocol PowerPlex® 16 BIO System for the PowerPlex® 16 BIO Allelic Ladder or Internal Lane Standard 600 BIO expiration date. All worksheets and paperwork associated with the quality control test will be maintained in a ring binder or properly labeled file.

NOTE: This quality control test can be done in conjunction with the quality control testing of the PCR-based kits, PAGE Plus™ gel solution, or AmpliTaq™ Gold DNA polymerase.

3.5 Standard Reference Material (SRM) 2391 Kit

The Standard Reference Material (SRM) 2391 kit is designed to assure that each step of the procedure for the DNA polymerase chain reaction (PCR) methodology is functioning properly and can be calibrated to the national standard provided by the National Institute of Standards and Technology (NIST). The SMR 2391 kit will be used on each applicable extraction and DNA typing system that is currently being used to analyze casework or Data Bank samples (e.g., stain extraction buffer with Microcons® , DNA IQ™ and Qiagen extractions).

3.5.1 Each laboratory conducting PCR analysis will verify its procedure using the SRM 2391 kit or samples traceable to a SRM kit annually.

3.5.2 Twelve samples are provided in the SRM 2391 kit (i.e., genomic 1-8, genomic GM09947A and GM09948, and GM09947A and GM09948 cells). Depending on the stage for which the sample is provided, the sample will be carried through the normal STR procedures addressed in the Forensic Biology procedure manual. Alternately, at least two NIST traceable samples can be processed in accordance with the procedures addressed in the Commonwealth of Virginia Division of Forensic Science Forensic Biology Section Procedure Manual, Section III, Fluorescent Detection PCR-Based STR DNA Protocol PowerPlex® 16 BIO System. Refer to appendix K for information on how to prepare NIST traceable samples.

3.5.3 Once the gel images have been sized, the expected types obtained and the controls typed appropriately, the laboratory's procedure will be considered to be functioning properly and calibrated to the national standard. **The quality control verification of the laboratory's PCR-based procedure does not need to be sized by an independent sizer**.

3.5.4 All worksheets and paperwork associated with the quality control test will be maintained in a ring binder or properly labeled file.

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